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ORIGINAL ARTICLE

The Relationship between Vitamin D Levels and Iron Deficiency and Anemia in Adults Applied for Periodic Medical Examination

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SUMMARY

Background: **Anemia and vitamin D deficiency are two important public health issues that may accompany many acute and chronic diseases. Several studies conducted in recent years have suggested that vitamin D deficiency is associated with anemia in healthy and patient populations. The aim of the study is to investigate the relationship between vitamin D deficiency and anemia.**

Methods: **The data of 9,590 adults aged 18 - 64, who applied for periodic medical examination to family medicine polyclinics of a training hospital between 2016 and 2018, were evaluated retrospectively. Individuals were classified into three groups as iron deficiency, iron deficiency anemia, and anemia; and 25 hydroxy vitamin D (25(OH)D) levels were classified into three groups as deficiency, insufficiency, and sufficiency. The groups were compared with respect to study parameters.**

Results: **Of the participants, 2,395 were male (25.0%) (mean age = 43.75 ± 13.43) and 7,195 (75.0%) were female (mean age = 42.93 ± 12.85). The number of anemic patients was 1,470 (15.3%) while the number of patients having no symptoms of anemia was 8,120 (84.7%). Serum hemoglobin (Hgb), iron, and ferritin levels were found to be significantly lower in the group with 25(OH)D deficiency than in the group of those with no deficiency. The mean 25(OH)D levels were observed to be significantly lower in those having anemia (17.4 ng/mL) than in those who do not (20.2 ng/mL), in those having iron deficiency (18.2 ng/mL) than in those who do not (20.5 ng/mL), and in those having iron deficiency anemia (16.6 ng/mL) than in those who do not (20.1 ng/mL) (all p-values are < 0.001).**

Conclusions: **The findings of this large study population, who live in a Mediterranean city which is sunny for 300 days of the year, indicate that 25(OH)D deficiency is significantly associated with iron deficiency and/or anemia.**
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KEY WORDS

vitamin D, vitamin D deficiency, anemia, iron deficiency, periodic medical examination

INTRODUCTION

Anemia and vitamin D deficiency are important public health problems that may accompany many acute and chronic diseases. While anemia affects approximately one in three people worldwide [1], vitamin D deficiency has become pandemic, affecting all age groups and all races [2,3].

The World Health Organization (WHO) defined anemia as serum hemoglobin (Hgb) levels of < 13 g/dL in men over 15 years of age, < 12 g/dL in women over 15 years

of age and in non-pregnant women, and < 11 g/dL in pregnant women [4]. Based on such clinical values, it is known that over 30% of the world's population is suffering anemia, more than half of which is due to iron deficiency [5].

Serum 25(OH)D is the best indicator for the amount of vitamin D existing in the body [6]. Despite a lack of consensus on the optimal level of serum 25(OH)D, it is considered sufficient when the level is above 30 ng/mL (75 nmol/L), insufficient when it is between 20 - 30 ng/mL (50 - 75 nmol/L), and deficient when it is below 20 ng/mL (50 nmol/L) [7]. Above all, the blood serum 25(OH)D level of > 150 ng/mL may lead to vitamin D intoxication [6].

In recent years, besides the well-known effects of vitamin D on bone and mineral metabolism, the other effects of vitamin D on many organs and tissues through its receptors (including bone marrow) have also been studied [8,9]. Furthermore, vitamin D deficiency is suggested to be associated with anemia in healthy and unhealthy population [10].

Several theories have been proposed to explain the relationship between vitamin D deficiency and anemia. Some of the theories suggested regarding vitamin D are that it promotes erythropoiesis by directly stimulating erythroid precursors, helps to store iron and reduce pro-inflammatory cytokines [11]**,** and suppresses hepcidin mRNA transcription by reducing pro-inflammatory cytokines [10]. Hepcidin is a hormone that is synthesized in the liver and its release is regulated by iron level, inflammation, infection, anemia or erythropoiesis [12]. Hepcidin has been shown to be inversely associated with iron absorption in healthy females and patients with iron deficiency [13].

Although there are many studies in the literature investigating the relationship between serum vitamin D levels and anemia in children and adolescents [14-17], elderly [18,19], women [20,21], chronic kidney patients [22,23] or different ethnic groups [24], there are few studies conducted with adult individuals with no obvious complaints, and the number of participants is usually low. This study aimed to investigate the relationship between 25(OH)D levels and anemia parameters in individuals who did not have any obvious complaints and who applied to the family medicine polyclinics of a training hospital in Antalya city, which is sunny for about 300 days of the year.

MATERIALS AND METHODS

Study population

The retrospective study included 9,590 people aged 18 - 64, who were admitted to the Family Medicine outpatient clinics of Saglik Bilimleri University (SBU) Antalya Training and Research Hospital (TRH) for periodic medical examination between 2016 and 2018, with the exclusion of presenting any symptoms which might cause iron deficiency and/or anemia. According to specifications of the WHO, anemia is defined as a Hgb level of < 13 g/dL in men and < 12 g/dL in non-pregnant women. Likewise, iron deficiency is defined as serum ferritin levels of < 15 ng/mL without having anemia and iron deficiency anemia is defined as serum ferritin levels of < 15 ng/mL accompanied by anemia (according to the WHO standards). Vitamin B12 < 200 pg/mL is considered as vitamin B12 deficiency without having anemia; whereas vitamin B12 deficiency anemia is considered as vitamin B12 < 200 pg/mL accompanied by anemia [25,26]. The patients were classified into five groups, namely iron deficiency, anemia, iron deficiency anemia, vitamin B12 deficiency and vitamin B12 deficiency anemia; whereas 25(OH)D levels were classified into three groups, namely deficiency, insufficiency, and sufficiency. The groups were compared with respect to study parameters. Due to the retrospective nature of the study, the applicants' information about whether they use vitamin D or not were not obtained.

Prior to the study, approval was obtained from Antalya TRH Clinical Research Ethics Committee (date: 16.08. 2018; decision no: 16/2), and the study was performed in compliance with the Declaration of Helsinki.

Data collection and laboratory measurements

In the collected blood samples, hemogram (complete blood count) parameters were analyzed by Beckman-Coulter LH780 hematology autoanalyzer, as a routine process of hospital laboratory. Likewise, iron, total iron binding capacity (TIBC), and unsaturated iron binding capacity (UIBC) tests were analyzed by Beckman-Coulter commercial kits on a Beckman-Coulter AU5800 auto-analyzer (Beckman-Coulter Inc., CA, USA), which is a conventional spectrophotometer. Ferritin, vitamin B12, and folic acid tests were performed by chemiluminescence method on Beckman-Coulter DxI 800 analyser (Beckman-Coulter Inc., CA, USA). In addition, 25(OH) D levels were measured by chemiluminescence method using Liasion (DiaSorin, MN, USA). All relevant data of the study population as to age, gender, available diagnostic information, hemogram parameters, iron, ferritin, TIBC, UIBC, folic acid, vitamin B12 and vitamin D levels were obtained from hospital records to data collection files.

Statistical analysis

The data collected were analyzed using IBM SPSS for Windows, v. 22.0 (IBM Corp., Armonk, NY). Categorical variables were compared using Pearson's chi-square analysis. The Sharipo-Wilk test was used to determine normality of the variables. The comparisons were made between the two groups using Student's *t*-test for normally distributed variables, and non-normal distributed variables were analyzed with the Mann-Whitney's *U*-test.

It was deemed necessary to use the Kruskal-Wallis test for the comparison of non-parametric variables between the groups and the Bonferroni-Dunn test as a post-hoc test for significant cases while One-Way ANOVA with

Table 1. Demographic features of patients and laboratory results.

Demographic features and laboratory results		Patients (n = 9,590)
Age (years)	Male	43.75 ± 13.43 (18 - 64)
	Female	42.93 ± 12.85 (18 - 64)
	Total	43.13 ± 13 (18 - 64)
Gender	Male	2,395 (25.0 %)
	Female	7,195 (75.0 %)
Anemia status	Anemic	1,470 (15.3%)
	Non-anemic	8,120 (84.7%)
Vitamin D status	Sufficient	1,786 (18.6%)
	Insufficient	2,938 (30.6%)
	Deficient	4,865 (50.7%)
Laboratory results	Hemoglobin (g/dL)	13.46 ± 1.62 (5 - 20.1)
	Hematocrit (%)	40.61 ± 4.38 (18.5 - 61)
	Iron (µg/dL)	78.11 ± 39.14 (1 - 541)
	UIBC (µg/dL)	290.17 ± 77.16 (6 - 663)
	TIBC (µg/dL)	368.28 ± 60.23 (44 - 827)
	Ferritin (ng/mL)	38.09 ± 40.64 (1 - 298.4)
	Vitamin B12 (pg/mL)	264.7 ± 155.05 (12 - 1495)
	Folic acid (ng/mL)	9.54 ± 3.91 (1.59 - 25.13)
	25(OH)D (ng/mL)	22.03 ± 13.71 (2.95 - 150)

Data are presented as n (%), mean ± SD (min - max). UIBC - unsaturated iron-binding capacity, TIBC - total iron-binding capacity.

Table 2. Comparison of anemic and non-anemic individuals.

Parameters	Anemic (n = 1,470)	Non-anemic (n = 8,120)	p-value
Age (years)	42.05 ± 12.25	43.33 ± 13.12	< 0.001
Gender (male/female)	141 (9.6)/1,329 (90.4)	2,254 (27.8)/5,866 (72.2)	< 0.001
Hemoglobin (g/dL)	10.98 ± 1.11	13.91 ± 1.25	< 0.001
Hematocrit (%)	34.26 ± 2.99	41.76 ± 3.52	< 0.001
Iron (µg/dL)	38 (1 - 312)	79 (1 - 541)	< 0.001
UIBC µg/dL)	358 (15 - 663)	274 (6 - 615)	< 0.001
TIBC (µg/dL)	399 (82 - 677)	358 (44 - 827)	< 0.001
Ferritin (ng/mL)	7 (1 - 297.7)	27.8 (1.9 - 298.4)	< 0.001
Vitamin B12 (pg/mL)	213 (43 - 1494)	229 (12 - 1495)	< 0.001
Folic acid (ng/mL)	8.51 (1.59 - 23.89)	8.86 (1.76 - 25.13)	0.003
25(OH)D (ng/mL)	17.4 (2.95 - 114)	20.18 (3.59 - 150)	< 0.001

Data are presented as n (%), mean ± SD, and median (min - max). Student's *t*-test, Mann-Whitney *U*-test, Pearson's Chi-square test. UIBC - unsaturated iron-binding capacity, TIBC - total iron-binding capacity.

post-hoc Tukey HSD test was used for parametric variables. Spearman's correlation coefficient was applied in order to identify the correlation between continuous variables. The association between vitamin D and the other study parameters was examined using multiple linear regression analysis. Data are expressed as n (%), mean ± standard deviation (SD) or median (min - max), as appropriate. Statistical significance was set at the 0.05 level.

Table 3. Comparison of the vitamin D levels and anemia status of patients.

Vitamin D status	Iron deficiency		Anemia		Iron deficiency anemia		Vitamin B12 deficiency		Vitamin B12 deficiency anemia	
	No n = 6,355	Yes n = 3,235	No n = 8,120	Yes n = 1,470	No n = 8,590	Yes n = 1,000	No n = 5,887	Yes n = 3,703	No n = 8,956	Yes n = 634
Vitamin D levels	20.5 3.6 - 150	18.2 2.9 - 150	20.2 3.6 - 150	17.4 2.9 - 114	20.1 3.6 - 150	16.6 2.9 - 114	21.2 3.6 - 150	17.8 2.9 - 150	20.03 3.6 - 150	15.7 2.9 - 87.6
p-value	< 0.001		< 0.001		< 0.001		< 0.001		< 0.001	
Vitamin D deficiency	3,030 (47.7) [a]	1,836 (56.8) [b]	3,987 (49.1) [a]	879 (59.8) [b]	4,234 (49.3) [a]	632 (63.2) [b]	2,662 (45.2) [a]	2,204 (59.5) [b]	4,446 (49.6) [a]	420 (66.2) [b]
Vitamin D insufficiency	2,057 (32.4) [a]	881 (27.2) [b]	2,554 (31.5) [a]	384 (26.1) [b]	2,699 (31.4) [a]	239 (23.9) [b]	1,894 (32.2) [a]	1,044 (28.2) [b]	2,788 (31.1) [a]	150 (23.7) [b]
Vitamin D sufficiency	1,268 (20.0) [a]	518 (16.0) [b]	1,579 (19.4) [a]	207 (14.1) [b]	1,657 (19.3) [a]	129 (12.9) [b]	1,331 (22.6) [a]	455 (12.3) [b]	1,722 (19.2) [a]	64 (10.1) [b]
p-value	< 0.001		< 0.001		< 0.001		< 0.001		< 0.001	

Data are presented as n (%) and median (min - max). Mann-Whitney *U*-test, Pearson's Chi-square test. Different lowercase letters in a row indicate statistically significant difference between groups.

Table 4. Comparison of iron deficient and anemic patients with control group as to their vitamin D status.

Vitamin D status	Iron deficiency n = 2,235	Anemia n = 1,470	Control group n = 5,885	p-value
Vitamin D levels	19 (4 - 150) [a]	17.4 (2.95 - 114) [b]	20.7 (3.59 - 150) [c]	< 0.001
Vitamin D deficiency	1,204 (53.9) [a]	879 (59.8) [b]	2,783 (47.3) [c]	< 0.001
Vitamin D insufficiency	642 (28.7) [a]	384 (26.1) [b]	1,912 (32.5) [c]	
Vitamin D sufficiency	389 (17.4) [a]	207 (14.1) [b]	1,190 (20.2) [c]	

Data are presented as n (%) and median (min - max). Kruskal-Wallis test, Pearson's Chi-square test. Different lowercase letters in a row indicate a statistically significant difference between groups.

Table 5. Correlation between the 25(OH)D levels of anemic and non-anemic patients and other factors.

Parameters	Anemic (n = 1,470)		Non-anemic (n = 8,120)	
	r	p	r	p
Hemoglobin (g/dL)	0.124	< 0.001	0.070	< 0.001
Hematocrit (%)	0.119	< 0.001	0.057	< 0.001
Iron (µg/dL)	0.149	< 0.001	0.079	< 0.001
UIBC µg/dL)	-0.149	< 0.001	-0.112	< 0.001
TIBC (µg/dL)	-0.128	< 0.001	-0.093	< 0.001
Ferritin (ng/mL)	0.123	< 0.001	0.067	< 0.001
Vitamin B12 (pg/mL)	0.168	< 0.001	0.216	< 0.001
Folic acid (ng/mL)	0.017	0.519	0.026	0.020

Spearman correlation test.
UIBC - unsaturated iron-binding capacity, TIBC - total iron-binding capacity.

Table 6. Comparison of patients as to their 25(OH)D values.

Parameters	25(OH)D < 20	25(OH)D = 20 - 30	25(OH)D > 30	p
Age (years)	42.29 ± 13.09 a	43.07 ± 12.92 b	45.54 ± 12.58 c	< 0.001
Gender (male/female)	1,037 (21.3)/3,829 (78.7)	888 (30.2)/2,050 (69.8)	470 (26.3)/1,316 (73.1)	< 0.001
Hemoglobin (g/dL)	13.3 ± 1.66 [a]	13.65 ± 1.6 [b]	13.61 ± 1.5 [b]	< 0.001
Hematocrit (%)	40.2 ± 4.49 [a]	41.07 ± 4.3 [b]	40.97 ± 4.06 [b]	< 0.001
Iron (µg/dL)	71 (1 - 541) [a]	77 (5 - 364) [b]	79 (1 - 374) [b]	< 0.001
UIBC (µg/dL)	292 (6 - 663) [a]	276 (7 - 592) [b]	271 (15 - 598) [c]	< 0.001
TIBC (µg/dL)	368 (82 - 827) [a]	359 (44 - 630) [b]	356 (82 - 646) [c]	< 0.001
Ferritin (ng/mL)	21.9 (1 - 297.7) [a]	27.1 (1 - 292.6) [b]	25.9 (1.4 - 298.4) [b]	< 0.001
Vitamin B12 (pg/mL)	209 (22 - 1495) [a]	234 (12 - 1448) [b]	267 (62 - 1446) [c]	< 0.001
Folic acid (ng/mL)	8.76 (1.59 - 25.13) [a]	8.63 (2.12 - 25.06) [a]	9.31 (1.88 - 24.68) [b]	< 0.001

Data are presented as n (%) and median (min - max). ANOVA with Tukey-HSD test, Kruskal-Wallis with Bonferroni-Dunn post-hoc test. Pearson Chi-square test. Different lowercase letters in a row indicate statistically significant difference between groups. UIBC - Unsaturated iron-binding capacity, TIBC - Total iron-binding capacity.

Table 7. Multiple logistic regression analysis of anemia.

Variables	OR (95% CI)	p-value
Age (years)	1.001 (0.996 - 1.007)	0.572
Male Gender	0.625 (0.507 - 0.771)	< 0.001
Iron (µg/dL)	0.967 (0.965 - 0.970)	< 0.001
TIBC (µg/dL)	1.005 (1.004 - 1.006)	< 0.001
Ferritin (ng/mL)	0.997 (0.995 - 0.999)	0.038
Vitamin B12 (pg/mL)	0.998 (0.997 - 1.001)	0.612
Folic acid (ng/mL)	1.001 (0.985 - 1.018)	0.899
25(OH)D (ng/mL)	0.990 (0.985 - 0.995)	< 0.001

RESULTS

A total of 9,590 people was included in the study, with 2,395 men (25.0%) (mean age = 43.75 ± 13.43) and 7,195 women (75.0%) (mean age = 42.93 ± 12.85). Of the participants, 1,470 (15.3%) were anemic, while 8,120 (84.7%) of them had no evidence of anemia. As to 25(OH)D levels, 1,786 people were found sufficient, 2,938 people were insufficient, and 4,865 people were deficient. Mean Hgb, hematocrit (Hct), iron, UIBC, TIBC, ferritin, vitamin B12, folic acid, and 25(OH)D levels were 13.46 ± 1.62 (g/dL), 40.61 ± 4.38 (%), 78.11 ± 39.14 (µg/dL), 290.17 ± 77.16 (µg/dL), 368.28 ± 60.23 (µg/dL), 38.09 ± 40.64 (ng/mL), 264.7 ± 155.05 (pg/mL), 9.54 ± 3.91 (ng/mL), and 22.03 ± 13.71 (ng/mL), respectively (Table 1).

When anemic and non-anemic patients were compared, mean 25(OH)D levels were observed to be significantly lower in anemic patients (17.4 ng/mL) than in non-anemic ones (20.18 ng/mL) ($p < 0.001$) (Table 2).

Participants were classified into five groups as iron deficiency, anemia, iron deficiency anemia, vitamin B12 deficiency, and vitamin B12 deficiency anemia and compared in terms of 25(OH)D levels. The mean level of 25(OH)D was significantly lower in patients with iron deficiency (18.2 ng/mL); anemia (17.4 ng/mL); iron deficiency anemia (16.6 ng/mL); vitamin B12 deficiency (17.8 ng/mL), and vitamin B12 deficiency anemia (15.7 ng/mL) than in those without (20.5 ng/mL), (20.2 ng/mL), (20.1 ng/mL), (21.2 ng/mL), and (20.03 ng/mL) respectively (all p-values are < 0.001). While the incidences of vitamin D deficiency and vitamin D insufficiency were significantly higher in people with iron deficiency, anemia, iron deficiency anemia, vitamin B12 deficiency, and vitamin B12 deficiency anemia, the incidence of vitamin D sufficiency was found to be significantly higher in those without (Table 3). When compared with the control group according to

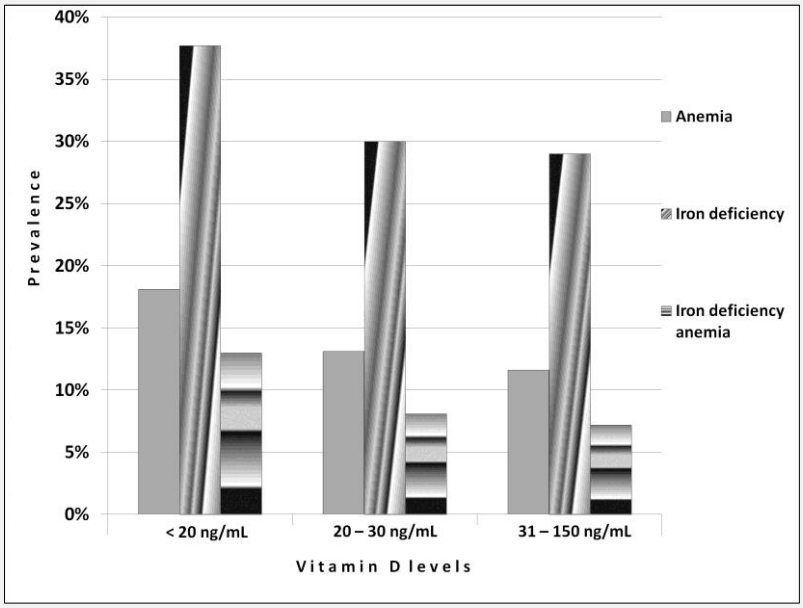

Figure 1. Prevalence of anemia, iron deficiency, and iron deficiency anemia that categorized by vitamin D levels.

mean serum vitamin D levels, it was observed that anemia group has the lowest mean level of serum 25(OH)D with 17.4 ng/mL, while the control group has the highest mean level of serum 25(OH)D with 20.7 ng/mL. Additionally, the rate of vitamin D deficiency was detected as the highest in the group of anemic patients with 59.8%, while the rate of vitamin D sufficiency patients were the highest in the control group with a percentage of 20.2% (Table 4).

The results showed that 25(OH)D was positively correlated with Hgb, Hct, serum iron, ferritin, vitamin B12, and folic acid whereas it was negatively correlated with UIBC and TIBC in anemic and non-anemic patients (Table 5).

The 25(OH)D levels obtained in the study were divided into three groups as sufficient (> 30 ng/mL), insufficient (20 - 30 ng/mL), and deficient (< 20 ng/mL), and their relationship with demographic characteristics and other laboratory findings were examined. The mean age, vitamin B12, and folic acid levels of the group with sufficient levels of 25(OH)D were found to be significantly higher than those who were 25(OH)D insufficient and deficient. On the other hand, the mean Hgb, Hct, iron, and ferritin levels were significantly lower in the 25(OH)D deficient group than in the 25(OH)D insufficient and sufficient group (Table 6). In addition, the prevalence of anemia, iron deficiency, and iron deficiency anemia was observed to be higher in the 25(OH) D deficient group (Figure 1).

The results of logistic regression analyses showed that gender, iron, TIBC, ferritin, and 25(OH)D were highly related factors for the development of anemia in patients. It was found that anemia was positively associated with TIBC (odds ratio, OR: 1.005; 95% CI: 1.004 - 1.006; $p < 0.001$) while it was negatively associated with male gender (OR: 0.625; 95% CI: 0.507 - 0.771; $p < 0.001$), iron (OR: 0.967; 95% CI: 0.965 - 0.970; $p < 0.001$), ferritin (OR: 0.997; 95% CI: 0.995 - 0.999; $p = 0.038$), and 25(OH)D (OR: 0.990; 95% CI: 0.985 - 0.995; $p < 0.001$) (Table 7).

DISCUSSION

The results of this study suggest that low serum 25(OH)D levels are associated with iron deficiency and/or anemia. These data support the previous studies that studied the association between vitamin D and its effects on erythropoiesis and hematopoietic system.

Kim et al. showed that in patients with end-stage renal disease [22] and Patel et al. showed that in patients with early-stage renal disease [23], there was a relationship with vitamin D deficiency and low hemoglobin levels and/or anemia. In our study, the exclusion of the patients with renal failure from the study revealed the association of vitamin D deficiency with low hemoglobin levels and/or anemia more clearly.

In a study by Lee et al. involving 2,526 healthy children and adolescents, the writers found that vitamin D deficiency was associated with the increased risk of anemia,

and iron deficiency anemia in particular [14]. Qader and Alkhateeb do not state that the children with vitamin D deficiency are prone to anemia, but that the children with iron deficiency are more prone to vitamin D deficiency, and that every child with iron deficiency anemia should also be evaluated in terms of vitamin D [16]. In our study, the increased prevalence of iron deficiency and/or anemia in 25(OH)D deficient patients and, likewise, the increased prevalence of vitamin D deficiency and insufficiency in patients with iron deficiency and iron deficiency anemia, supported both studies mentioned above.

In a study conducted by Nikooyeh and Neyestani with 937 children aged 9 - 12 years, low vitamin D levels were associated with increased risk of anemia independent of age, gender and body mass index. In comparison to the vitamin D-sufficient children in a similar age group, the vitamin D-deficient children were observed to develop three times more anemia, and anemia risk increased in patients with serum 25(OH)D levels of < 44 nmol/L (17.6 ng/mL) [15]. In another study conducted with 10,410 healthy American children and adolescents aged 1 - 21, 25(OH)D deficiency was associated with increased risk of anemia, and the threshold level of 25(OH)D (< 12 ng/mL) for low Hgb counts was found to be lower in black children than in white children [17].

There are also studies in the literature that do not support the relationship between vitamin D level and anemia. For example, Madar et al. classified the participants with low vitamin D levels into three groups in a randomized, double-blind, placebo-controlled study of different ethnic groups (South Asia, Middle East, and Africa) living in Norway. Group 1 was given 10 µg of vitamin D3, while Group 2 was given 25 µg of vitamin D3 daily for 16 weeks, after which both groups were compared with the placebo group. The study showed that vitamin D3 supplementation did not significantly affect Hgb levels and other parameters indicating the iron status [24]. In a retrospective study by Öztürk et al. conducted with 302 patients, the writers classified the patients into four groups according to vitamin D levels and indicated that there was no significant difference in Hgb levels between the groups [27].

In one of the few studies showing the direct relationship between vitamin B12 deficiency and vitamin D deficiency, Massironi et al. found a significant correlation between serum vitamin B12 levels and 25(OH)D levels in patients with autoimmune atrophic gastritis [28]. Similar to the study of Massironi, in our study serum 25(OH)D levels were found to be significantly lower in the group with vitamin B12 deficiency. It was thought to be caused by malnutrition and malabsorption, which could lead to deficiency of both vitamins.

As suggested by Qader et al., extensive study groups and cohort studies are needed to clarify this relationship [16]. No matter how large our study group is, there are still some limitations in our study that affect the results. First, in our study, the patients were evaluated cross sectionally according to the results of a single examination. Next, as it is a retrospective study, no data could be gathered on whether all of the participants received vitamin D and/or iron replacement.

However, unlike other studies showing the relationship between vitamin D and iron deficiency and/or anemia, one of the strengths of our study is that the data of almost ten thousand healthy individuals were evaluated. The second one is that the patients with other diagnoses that may be the cause of anemia such as anemia of inflammation or anemia related to genetic factors like sickle cell disease and thalassemia were all excluded. As a result, vitamin D deficiency is associated with iron deficiency and/or anemia. Vitamin D may prevent the development of anemia by supporting erythropoiesis. Therefore, all patients detected with vitamin D deficiency in the daily outpatient clinic should also be evaluated for iron deficiency and/or anemia, and those who need should be given replacement therapy.

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Declaration of Interest:
Authors have nothing to declare.

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